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THE TOLERANCE OF SOYBEANS TO ATRAZINE, 2-CHLORO-4
(ETHYLAMINO)-6-(ISOPROPYL AMINO)-S-TRIAZINE

by

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A MASTER'S THESIS

submitted in partial fulfillment of the

requirements for the degree

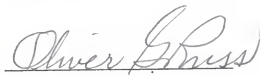
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Soybean Tolerance to Atrazine

Abstract. Corn (Zea mays L.) and Soybeans (Glycine max L. Merrill) frequently are grown in rotation in the Midwest. Soybeans were planted immediately after applying and incorporating 2-chloro-4-(ethylamino)-6-(isopropylamino)-s-triazine (atrazine) at rates up to 2.24 kg/ha. Yield reductions occurred about 30% of the time a 1.68 and 2.24 kg/ha rates of atrazine. Plant populations were decreased in all cases where 2.24 kg/ha had been applied, and frequent stand reductions were observed at the 1.12 and 1.68 kg/ha rates. Soybean plants were able to compensate for stand reductions by setting more pods on the remaining plants. Atrazine toxicity appears to be linked to the microenvironment. High soil moisture conditions at planting and early planting tended to reduce atrazine toxicity. High moisture at planting and during the early susceptible stage of plant growth may dilute the atrazine.

Soybean seed collected from plots treated with 0.0 kg and 2.24 kg/ha of atrazine was graded into large and small seeds and planted the following year where 2.24 kg/ha atrazine had been applied preplant incorporated. No significant differences were observed in yield, protein, or seed weight for either previous atrazine treatment or seed size. However, plant population was significantly higher in 1972 and 1973 for large as compared to small seeds.

Introduction

Atrazine commonly is used alone or in combination with other herbicides on corn in the Midwest, as a preemergent, preplant incorporated, or early post treatment. Since soybeans and corn frequently are grown in rotation and soybeans are marginally tolerant to atrazine, its use has presented residue problems the same year and the year after application.

Corn can metabolize atrazine into products which are not injurious (15, 18). Soybeans, which show some tolerance, can metabolize atrazine but at a slower rate (14).

The effect of atrazine on soybeans the year after application has been previously investigated (3, 9). Soil factors, temperature (6, 12, 17), moisture (7) pH (15), and organic matter (2, 12, 15), and previous application rates (2, 4) will influence the amount of atrazine residue in succeeding years. Little work has been done to determine the effect of atrazine on soybeans that are planted in the year of herbicide application. Insects, disease, hail, or heavy rain may make it desirable to plant soybeans after a corn crop has been injured or destroyed or not planted.

Environmental conditions between the time atrazine is applied and when soybeans are planted affect the amount of atrazine residue. McCormick and Hilthold (13) found that for each 10 C rise in temperature from 10 to 30 C decomposition of atrazine is doubled. Burnside (5) found the average temperature below the plow layer to be 12 C, which helps to explain the persistence of herbicides under field conditions.

The amount of rainfall and velocity of leaching are important factors in decreased toxicity. Atrazine is not leached rapidly since its solubility is 33 ppm, but Burnside et al. 6 (3) found that atrazine leached to the 30 to 45 cm soil depth after four months and to the 45 to 61 cm depth or more after 16 months.

Behrens and Carwent (3) found a slight reduction in soybean yield the year after four annual atrazine applications of 3.4 kg/ha.

Birk and Rhoadhouse (4) found that 8.1 percent of the atrazine applied remained one year after treatment.

Harris, Woolson, and Hummus (10) found that increasing soil organic matter, depth of placement, and decreasing temperature tended to make a herbicide more persistent.

Repeated use of a given herbicide can result in an accumulation of phytotoxic residues if there is not continual removal or inactivation (2). Klingman (11) states that removal or inactivation can be stimulated by adsorption on the soil colloid, chemical or microbial decomposition, photodecomposition, and volatilization.

Anderson (1) in greenhouse work with 2700 strains of soybeans found a wide variation in tolerance to atrazine. He found that tolerance generally increased as seed size increased. Regression analysis indicated that 80% of the variation in response was attributed to variation in seed size. He suggested the possibilities of minimizing soybean injury from atrazine and other herbicides by planting large seeds and suggested field studies should be done.

Seeds of susceptible plants usually germinate, but the plants die shortly after emergence due to uptake of atrazine through the roots. The first symptom of injury is marginal chlorosis. If the soybean plant

escapes early injury, it probably will survive. Small sublethal amounts of atrazine have been found to stimulate growth of soybeans (7).

This study was initiated to determine if soybeans could be grown where atrazine had been applied in the same year prior to soybean planting and if seed size influenced the tolerance of soybeans to atrazine.

Methods and Materials

The study was conducted at Manhattan, Kansas, at the Ashland Unit of the Agronomy Research Farm in 1971, 1972, and 1973 on a Pachic Haplustolls soil (Muir silt loam) containing 2.5 percent organic matter and having a slope of less than 1.0 percent.

A split plot design was used with five herbicide treatments and three replications and was planted on three dates each year: May 15, June 1, June 15. The plots were 9.1 m long and 3.0 m wide. Four 0.76 m rows were planted and the center two rows were harvested.

Atrazine was applied preplant incorporated at 0.0, 0.56, 1.12, 1.68, and 2.24 kg/ha. The atrazine was applied lengthwise in the plots and tandem disked approximately 10 cm deep, springtoothed, and tine-toothed. Soybeans were planted immediately after herbicide incorporation. To control grassy weeds, 1.12 kg/ha a, a, a, trifluoro-2, 6 dinitro-N, N-dipropyl-p-toluidine (trifluralin) was applied and incorporated with the atrazine. The herbicides were applied in 7.5 liters of water per hectare. The plots were not cultivated.

In the seed size section large and small seeds were planted where 2.24 kg/ha of atrazine was applied preplant incorporated as in

the other section at the same location. The small seeds weighed about 9.5 g/100 and the large seeds about 17.9 g/100. The planting date was June 1, 1971, 1972, and 1973. The plots were 9.1 m long and 30 m wide. Four 0.76 m rows were planted and the center rows harvested. Tri-fluralin was applied as in the rate study.

Yield, height, seed weight, plant count, and percent protein were measured each year. Height was the average of 10 random soybean plants per plot. Seed weight was recorded as grams/100 seeds. Plant count was recorded as plants/meter and protein was determined using the Kjeldahl procedure.

Results and Discussion

Yield. No significant yield reductions were found in 1971 on any date due to atrazine (Figure 1). In 1972 no significant reduction at any rate was recorded for Date I. Yield was reduced at the 1.68 and 2.24 kg rates in Date II and the 2.24 kg rate, at Date III. 1973 yields showed a reduction at the upper rate in Date I, Date II at the 3 higher rates, and Date III at the 2 upper rates. The Date X rate interaction was significant at the 1.0% level.

Plant population. In 1971 plant population was decreased at the 3 upper rates of atrazine (Figure 2). A significant decrease was observed at the 2.24 kg rate on all 3 dates and at the 1.68 kg rate on Date II. In 1972 and 1973 when the three dates were combined a significant reduction was observed at the 3 upper rates (Figure 2). When data from all three years were combined a significant reduction occurred at the 1.12 kg rate and above of atrazine. Although significant population

decreases were noted at the 1.12 kg rate and above, significant yield reductions usually were not observed until the 1.68 kg rate or 2.24 kg rate was applied. The date, year x date, rate, and year x date x rate interactions were significant at the .01% level. The soybean plant compensates for reduced stand by branching. LeBaron (12) found soybeans showed little or no correlation between early stand evaluations and yield.

Protein content. Previous research (8, 17) has indicated that application of triazines may influence protein content. However, in this study, protein content was not statistically different in any year. The means for 1971-1973 were 0.0, 38.7%; 0.56, 38.9%; 1.12, 38.5%; 1.68, 38.1% and 2.24, 39.1%

Seed weight. Seed weight ranged from 12.3 to 13.1 grams/100 seeds for atrazine rate means. Seeds from 1.68 and 2.24 kg/ha rates averaged slightly heavier, but the differences were not significant. If real, these higher weights may be attributed to the lower plant population, since there was less competition for light, nutrients, and moisture.

Height. Significant height differences were found in all years (Figure 3). These differences probably contributed to yield reductions by decreasing the total number of seeds per plant; also, the later the planting the shorter the plants.

Rainfall and temperature. Rainfall and temperature appear to influence atrazine injury to soybeans. In 1971 with heavy rains throughout the spring and summer, no yield reductions occurred. In 1972 yields were

not reduced on Date I even though little rain fell the week prior to planting (Table 1) which may be explained by the previous excellent moisture conditions present early that summer and the cool temperatures. Light rain fell before planting on Date II but none after. Injury occurred at the two higher rates of atrazine. Heavy rain fell before Date III and injury occurred only at the 2.24 kg rate of atrazine. In 1973 light rain fell before Dates I and II. Significant injury occurred at the 2.24 kg on both dates and the 1.12 and 1.68 kg rate on Date II. Good soil moisture conditions early in the spring of 1973 may have reduced the injury on Date I. Above average temperatures and windy weather were encountered after planting of Date II. Only light rain fell before planting Date III and injury occurred at the 1.68 and 2.24 kg rates. When rain occurred the week before planting or previous rains had brought the soil to field capacity, little atrazine injury occurred. Field capacity is the amount of water retained in the soil after gravitational drainage of excess water. The later the date of planting the more frequently injury occurred. This may have been due to higher temperatures, increasing transpiration and water uptake.

The more soil moisture present at planting, perhaps the more diluted the atrazine is in the soil solution. The more diluted the atrazine is, the more moisture the soybean plant must take up for toxic amounts of atrazine to be taken up. High temperatures and wind enhance transpiration and as the plant takes up moisture more atrazine is taken up. Date II in 1973 was followed by higher temperatures and wind; plant population and yield were both significantly reduced at the 1.12 kg/ha rate and above.

Vostrál et al. (20) showed that the amount of atrazine absorbed by soybeans increased with increases in herbicide concentration, absorption time, and root temperatures. Rieder et al. (17) found a direct relationship between uptake and concentration and noted that increasing the temperature from 10 to 30 C increased uptake of herbicides. After 48 hours soybeans seeds had taken up 65% of the atrazine in solution.

Early planting generally avoided atrazine injury. Early in the year good moisture conditions are usually present even if little rain falls immediately prior to planting. The cooler weather normally occurring early in the summer lowers transpiration and water uptake. After the plant roots have grown below the depth of incorporation, injury is not likely to occur. The soybean plants are most susceptible in early stages of growth when their roots are in the layer of soil where the atrazine is concentrated.

The seed size study indicated there were no significant differences for yield, protein content, seed weight, or height. In 1971 there were no significant differences in plant population. In 1972 and 1973 the large seed produced significantly more plants per meter of row. (Figure 4)

Germination was determined in 1972 and 1973 (Table 3). In 1972 large seeds germinated significantly better than small seeds.

Although significant differences in plant population were found these differences were not reflected in yield (Table 4). The soybean plant apparently compensated by setting more pods on the remaining plants.

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Table 1. Rainfall in the week prior to planting of Clarke 63 soybeans at Manhattan, Kansas.

Year	Planting Date		
	I	II	III
	cm		
1971	5.82	5.21	6.76
1972	1.65	1.40	5.12
1973	.58	.43	2.38

Table 2. Correlation between rainfall the week before planting and yield of Clark 63 soybeans.

	Atrazine Rate				
	0.00	0.56	1.12	1.68	2.24
r	.61	.78	.77	.81	.82

Table 3. Percent germination of Clark 63 Soybeans as affected by seed size, where 2.24 kg/ha of atrazine was applied.

Year	Seed Size	
	Small	Large
1972	84 ^a *	91 ^b
1973	94 ^a	89 ^a
Ave.	89	90

* Values within the same row with different letters are significantly different at the 5% level according to Duncan's Multiple Range Test.

Table 4. Yield of Clark 63 soybeans planted where 2.24 kg of atrazine was applied preplant incorporated.

Treatment		1971	Yield 1972	1973	\bar{X}	1971	1972	1973	\bar{X}
Small	2.24	2108.7	2151.5	1123.9	10.5	7.9	1.3		
Large	2.24	2215.7	2178.3	1536.0	11.2	10.8	2.3		
LSD	(.05)	663.6	N.S. *	492.4	3.6	N.S.	.7		

* N.S. No significant differences within column at the 5% level.

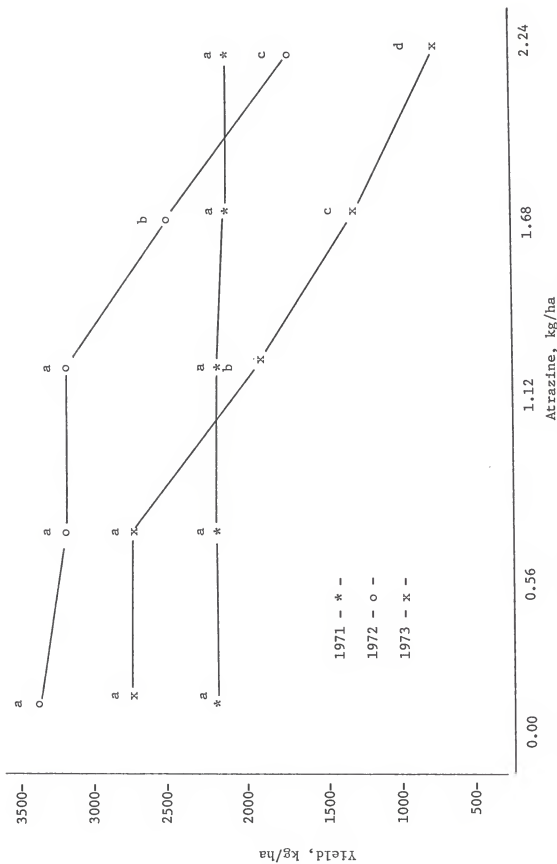


Figure 1. Yield of Clark 63 soybeans in 1971 to 1973 where atrazine was applied preplant incorporated. Points bearing the same letter within a year are not significantly different at the 5% level according to Duncan's Multiple Range Test.

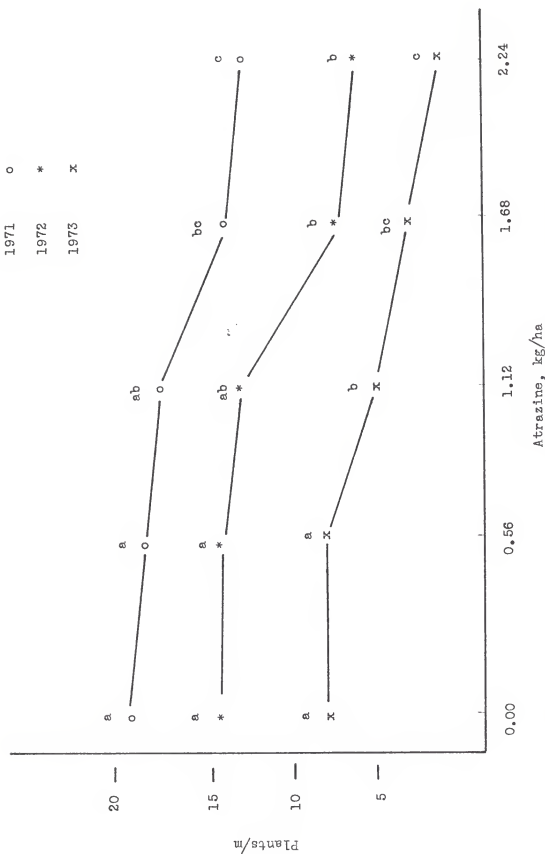


Figure 2. Plant population of Clark 63 soybeans in 1971 to 1973 planted where atrazine was applied preplant incorporated. Points bearing the same letter are not significantly different at the 5% level according to Duncan's Multiple Range Test.

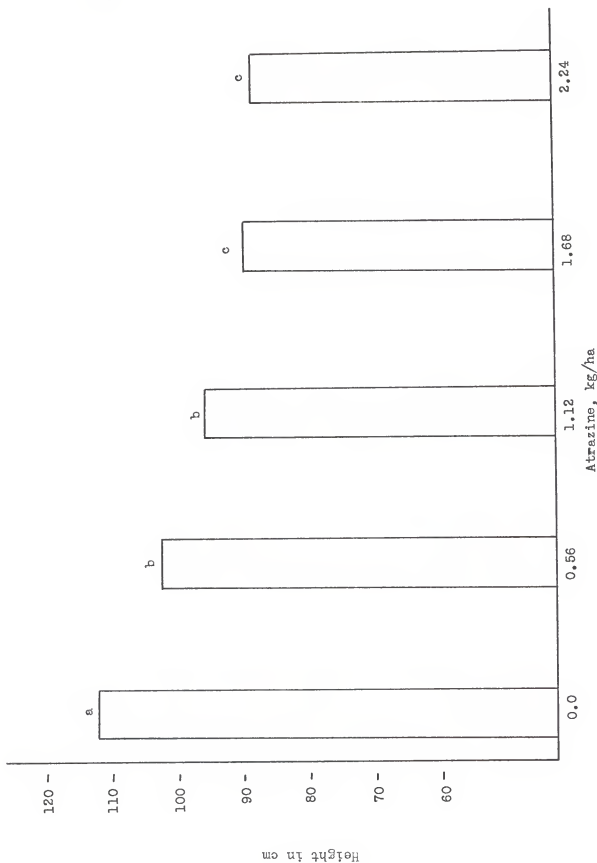


Fig. 3. Height of Clark 63 Soybeans in 1971-73 where atrazine was applied preplant incorporated. Differences significant at the 5% level according to Duncan's Multiple Range Test.

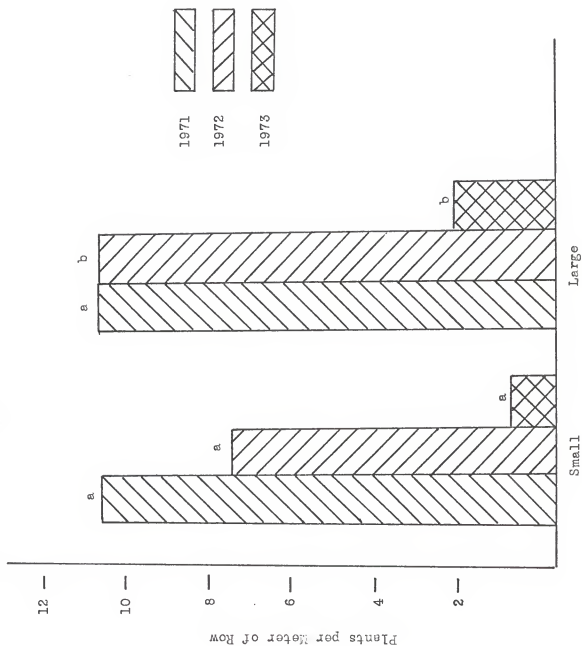


Figure 4. Plant population of Clark 63 soybeans in 1971 to 1973 where 2.24 kg of atrazine was preplant incorporated. Similar columns bearing the same letter are not statistically different at the 5% level according to Duncan's Multiple Range Test.

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APPENDIX

Table 1. Yield of Clark 63 soybeans planted where atrazine had been applied preplant incorporated with 3 dates of planting.

Rate of Atrazine	Year												
	1971 Date				1972 Date				1973 Date				
	I	II	III	Mean	I	II	III	Mean	I	II	III	Mean	Rate Mean

^a Values in the same column followed by the same letter are not significantly different at the 5% level according to Duncan's Multiple Range Test.

Table 2. Plant population of Clark 63 soybeans planted where atrazine had been applied preplant incorporated with 3 dates of planting.

Rate of Atrazine	Year												Rate Mean
	1971				1972				1973				
	I	II	III	Mean	I	II	III	Mean	I	II	III	Mean	
kg/ha	p1s/m												
0.00	16.8 ^a	18.0 ^a	19.8 ^a	18.3 ^a	17.1 ^a	14.9 ^a	11.6 ^a	14.0 ^a	11.6	4.9	5.8 ^a	7.3 ^a	13.1 ^a
0.56	14.6 ^{ab}	18.3 ^a	19.2 ^a	17.4 ^a	15.8 ^a	13.1 ^{ab}	12.5 ^a	14.0 ^a	8.8 ^a	6.7	5.8 ^a	7.0 ^a	12.8 ^a
1.12	15.5 ^{ab}	15.2 ^{ab}	15.2 ^{ab}	15.2 ^{ab}	12.5 ^b	9.8 ^b	11.9 ^a	11.3 ^{ab}	8.5 ^a	2.1 ^c	4.0	4.9 ^b	10.4 ^{ab}
1.68	13.4 ^{ab}	11.0 ^c	13.4 ^{ab}	12.5 ^{bc}	11.0 ^b	4.6 ^c	8.2 ^b	7.9 ^b	5.2 ^b	1.5 ^c	2.4 ^b	3.1 ^{bc}	7.9 ^{bc}
2.24	10.1 ^b	11.3 ^{bc}	9.1 ^b	10.0 ^c	7.4 ^c	1.2 ^c	5.2 ^c	5.0 ^b	2.1 ^c	.6 ^c	2.1 ^b	1.5 ^c	6.1 ^c
Mean	14.0	14.9	15.2	14.6	13.7	8.5	9.8	10.7	7.3	3.1	4.0	4.9	10.1
LSD	5.8	4.0	8.2	4.6	3.1	4.3	2.4	2.4	1.8	2.1	1.5	1.8	1.5

^a Values in the same columns followed by the same letter are not significantly different at the 5% level according to Duncan's Multiple Range Test.

Table 3. Analysis of Variance--Atrazine Rate Study

Source	df	Mean Square			
		yield	plants/meter	protein	height
Replicates	2	5.31	39.27	14.63	329.03
Years	2	69.45 ^{**}	12331.69 ^{**}	102.57 [*]	1968.23 ^{**}
Error (a)	4	2.40 ^{**}	25.19	12.66	67.55
Dates	2	29.16 ^{**}	922.38 ^{**}	10.62	7950.54 ^{**}
Year x Date	4	27.34 ^{**}	461.0. ^{**}	28.41	1236.80 ^{**}
Error (b)	12	1.15	23.71	17.64	59.49
Rate	4	43.58 ^{**}	2952.39 ^{**}	27.30	2696.77 ^{**}
Year x Rate	8	11.20 ^{**}	36.94	15.84	108.35
Date x Rate	8	5.21 ^{**}	64.18	8.70	111.25
Year x Date x Rate	16	2.38 [*]	129.41 ^{**}	12.21	102.72
Error (c)	72	1.17	39.40	13.64	123.29

^{**} Indicates significance at the 1% level.

Table 4. Analysis of Variance--Soybean Seed Source.

Source	df	Meter Square			
		yield	plants/meter	protein	height
Replicates	2	28404.6	36.15	39.35	282.53
Years	2	373279.3	2008.00**	651.96*	5618.53**
Error A	4	6728.6	23.74	47.87	197.61
Seed Size	1	77130.5	4.99	669.08*	1736.11
Atrazine Rate	1	993.5	9.82	.22	13.44
Year x Seed Size	2	13863.6	169.74	658.97	3963.53**
Year x Atra- zine Rate	2	903.2	3.00	0.59	7.19
Seed Size x Atrazine Rate	1	180.6	1.96	0.40	11.11
Year x Size x Rate	2	316.1	1.11	0.09	19.19
Error B	18	19011.7	55.11	99.90	552.14

** Indicates significance at the 1% level.

* Indicates significance at the 5% level.

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Soybean Tolerance to Atrazine

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Abstract. Corn (Zea mays) and soybeans (Glycine max L.) are frequently grown in rotation in the Midwest. Soybeans were planted immediately after applying and incorporating 2-chloro-4-(ethylamino)-6-(isopropyl-amino)-s-triazine (atrazine) at rates up to 2.24 kg/ha. Yield reductions occurred about 30% of the time at 1.68 and 2.24 kg/ha rates of atrazine. Plant populations were decreased in all cases where 2.24 kg/ha had been applied, and frequent stand reductions were observed at the 1.12 and 1.68 kg/ha rates. Soybean plants were able to compensate for stand reductions by branching. Atrazine toxicity appears to be linked to the microenvironment. High soil moisture conditions at planting and early planting tended to reduce atrazine toxicity. High moisture at planting and in the early susceptible stage of plant growth may dilute the atrazine. Lower temperatures early in the season reduced transpiration and may reduce total atrazine uptake.

Soybean seed collected from plots that had been treated with 0.0 kg and 2.24 kg/ha of atrazine was graded into large and small seeds and the next year planted where 2.24 kg/ha of atrazine had been applied preplant incorporated. No significant differences were observed in yield, protein, or seed weight for either previous atrazine treatment or seed size. However, a significant increase in plant population was noted in 1972 and 1973 between large and small seeds.